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L3: Entry 3 of 3 File: USPT Aug 23, 1994

US-PAT-NO: 5340738
DOCUMENT-IDENTIFIER: US 5340738 A

TITLE: Modified prokaryotic glucose isomerase enzymes with altered pH activity profiles

DATE-ISSUED: August 23, 1994

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Lambeir; Anne-Marie	Heverlee	N/A	N/A	BEX
Lasters; Ignace	Antwerp	N/A	N/A	BEX
Mrabet; Nadir	Hoeilaart	N/A	N/A	BEX
Quax; Wilhelmus J.	Voorschoten	N/A	N/A	NLX
Van der Laan; Jan M.	Groningen	N/A	N/A	NLX
Misset; Onno	Delft	N/A	N/A	NLX

US-CL-CURRENT: 435/234; 435/252.3, 435/252.33, 435/488, 435/69.1, 536/23.2

CLAIMS:

We claim:
1. A substantially pure, recombinantly-produced, modified procaryotic glucose isomerase containing a modification wherein at least one and no more than four amino acid residues from the corresponding naturally-ocurring glucose isomerase within a sphere of 15 A of a bivalent metal cation coordination site is replaced by a more positively charged amino acid, which modified glucose isomerase exhibits an altered pH/activity profile wherein at least the acidic part of the pH/activity profile is shifted to a lower pH value and wherein said modified glucose isomerase retains glucose isomerase activity.
2. The modified glucose isomerase of claim 1 wherein the corresponding naturally-occurring glucose isomerase is derived from a microorganism of the order Actinomycetales.
3. The modified glucose isomerase of claim 2 wherein the Actinomycetales microorganism is Actinoplanes missouriensis.
4. The modified glucose isomerase of claim 3 wherein at least one replaced amino acid of the corresponding naturally-occurring glucose isomerase replaced is selected from the group consisting of:
Ala5, Phe11, Leu15, Trp20, Gln21, Ala25, Phe26, Asp28, Ala29, Gly47, Tyr49, Thr52, Phe53, His54, Asp56, Asp57, Phe61, Ile85, Met88, Phe94, Thr95, Phe104, Gln122, Thr133, Leu134, Val135, Ala143, Tyr145, Tyr158, Asn163, Ser169, Glu181, Asn185, Glu186, Gly189, Ile191, Pro194, His198, Gln204, Leu211, Phe212, Asn215, Glu217, Thr218, His220, Glu221, Gln222, Ser224, Asn225, Leu226, Phe228, Thr229, Gly231, Leu236, His238, His243, Asp245, Asn247, His250, Phe254, Asp255, Gln256, Asp257, Leu258, Val259, Phe260, His262, Leu271, Tyr285, Asp286, His290, Asp292, Tyr293, Thr298, Glu299, Trp305, Ala310, Met314, Val380, and Asn383,
said positions referring to the wildtype glucose isomerase of Actinoplanes missouriensis, or
wherein said amino acid replaced is an amino acid at a corresponding position in a homologous glucose isomerase.
5. The modified glucose isomerase of claim 4 wherein said replaced amino acid is selected from the group consisting of:
Ala5, Leu15, Gln21, Ala25, Phe26, Asp28, Ala29, Gly47, Tyr49, Thr52, Asp56, Phe61,

Ile85, Met88, Thr95, Phe104, Gln122, Thr133, Leu134, Ala143, Tyr145, Tyr158, Asn163, Ser169, Asn185, Gly189, Ile191, Pro194, His198, Gln204, Leu211, Phe212, Thr218, Gln222, Ser224, Asn225, Leu226, Phe228, Thr229, Gly231, Leu236, His238, His243, His250, Phe254, Gln256, Asp257, Leu258, Val259, His262, Leu271, Tyr285, Asp286, His290, Tyr293, Thr298, Glu299, Trp305, Ala310, Met314, Val380, Asn383, said positions referring to the wildtype glucose isomerase of Actinoplanes missouriensis, or
wherein said amino acid replaced is an amino acid at a corresponding position in a homologous glucose isomerase.

6. The modified glucose isomerase of claim 5 wherein said replaced amino acid residue is selected from the group consisting of:
Ala25, Gly47, Tyr49, Thr52, Phe61, Ile85, Thr95, Gln122, Thr133, Tyr145, Tyr158, Gly189, Ile191, Gln204, Thr218, Gln222, Leu226, Thr229, Gly231, Leu236, Gln256, Leu258, Tyr293 and Val380,
said positions referring to the wildtype glucose isomerase of Actinoplanes missouriensis, or
wherein said amino acid replaced is an amino acid at a corresponding position in a homologous glucose isomerase.

7. The modified glucose isomerase of claim 5 wherein said replaced amino acid residue is selected from the group consisting of:
A25K, D57N, F61K, F94R, E186Q, Q204K, F254K, D255N, L258K, and H290N,
said positions referring to the wildtype glucose isomerase of Actinoplanes missouriensis, or
wherein said amino acid replaced is an amino acid at a corresponding position in a homologous glucose isomerase.

8. A method to produce a modified procaryotic gluose isomerase of claim 1, which process comprises:
mutating a DNA sequence encoding a wildtype glucose isomerase at selected nucleotide positions;
cloning the mutated sequence into an expression vector in such a manner that the DNA sequence can be expressed;
transforming a host organism or cell with the vector;
culturing the host organism or cell; and
isolating the modified glucose isomerase from the culture.

9. A process for producing the modified glucose isomerase of claim 1 which comprises culturing a host organism transformed with a mutated glucose isomerase DNA cloned into an expression vector in such a manner that the DNA sequence can be expressed under conditions which favor expression and
isolating the modified glucose isomerase from the culture.

10. The modified glucose isomerase of claim 7 wherein said amino acid replacement is L258K.

11. The modified glucose isomerase of claim 7 wherein said amino acid replacement is F61K.

12. The modified glucose isomerase of claim 7 wherein said amino acid replacement is E186Q.

13. The modified glucose isomerase of claim 7 wherein said amino acid replacement is Q204K.

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End of Result Set

Generate Collection

L3: Entry 3 of 3 File: USPT Aug 23, 1994

US-PAT-NO: 5340738
DOCUMENT-IDENTIFIER: US 5340738 A

TITLE: Modified prokaryotic glucose isomerase enzymes with altered pH activity profiles

DATE-ISSUED: August 23, 1994

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Lambeir; Anne-Marie	Heverlee	N/A	N/A	BEX
Lasters; Ignace	Antwerp	N/A	N/A	BEX
Mrabet; Nadir	Hoeilaart	N/A	N/A	BEX
Quax; Wilhelmus J.	Voorschoten	N/A	N/A	NLX
Van der Laan; Jan M.	Groningen	N/A	N/A	NLX
Misset; Onno	Delft	N/A	N/A	NLX

ASSIGNEE INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY	TYPE CODE
Gist-brocades, N.V.	Delft	N/A	N/A	NLX	03
Plant Genetic Systems, N.V.	Brussels	N/A	N/A	BEX	03

APPL-NO: 7/ 637399
DATE FILED: January 4, 1991

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	APPL-DATE
EP	90200030.6	January 4, 1990
EP	90200037.1	January 4, 1990

INT-CL: [5] C12M 15/61, C12M 9/92, C12M 15/70, C12M 15/74
US-CL-ISSUED: 435/234; 435/69.1, 435/172.3, 435/252.3, 435/252.33, 536/23.2, 935/10, 935/14, 935/60, 935/72, 935/75
US-CL-CURRENT: 435/234; 435/252.3, 435/252.33, 435/488, 435/69.1, 536/23.2
FIELD-OF-SEARCH: 435/234, 435/69.1, 435/172.3, 435/252.3, 435/252.33, 435/320.1, 536/23.2

REF-CITED:

U.S. PATENT DOCUMENTS

Search Selected Search ALL

	PAT-NO	ISSUE-DATE	PATENTEE-NAME	US-CL
☐	4609625	September 1986	Keyes et al.	435/176
☐	4894331	June 1990	Ratzkin et al.	435/94
☐	5041378	August 1991	Drammond et al.	435/234

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FOREIGN-PAT-NO	PUBN-DATE	COUNTRY
90-0196	January 1990	WOX

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Russel, A. J., et al., 1987, Journal of Molecular Biology, 193: 803-813.
Thomas, P. G., et al., 1985 Nature, 318: 375-376.
Collyer, L. A., et al., Mar. 1990, Journal of Molecular Biology, 212(1): 211-235.
Henrick, K., et al., 1984, Journal of Molecular Biology, 208(1): 129-157.
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ART-UNIT: 184
PRIMARY-EXAMINER: Wax; Robert A.
ASSISTANT-EXAMINER: Moore; William M.
ATTY-AGENT-FIRM: Morrison & Foerster

ABSTRACT:

A method for selecting amino acid residues is disclosed which upon replacement will give rise to an enzyme with an altered pH optimum. The method is specific for metalloenzymes which are inactivated at low pH due to the dissociation of the metal ions. The method is based on altering the pK.sub.a of the metal coordinating ligands or altering the K.sub.ass for the metal binding. New glucose isomerases with an altered pH optimum are provided according to this method. These altered properties enable starch degradation to be performed at lower pH values.

13 Claims, 24 Drawing figures

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L3: Entry 2 of 3 File: USPT Dec 27, 1994

US-PAT-NO: 5376536
DOCUMENT-IDENTIFIER: US 5376536 A

TITLE: Glucose isomerase enzymes and their use

DATE-ISSUED: December 27, 1994

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Quax; Wilhemus J.	VB Voorschoten	N/A	N/A	NLX
Luiten; Rudolf G. M.	KR Leiden	N/A	N/A	NLX
Schuurhuizen; Paul W.	NT Delft	N/A	N/A	NLX
Mrabet; Nadir	Hoeilaart	N/A	N/A	BEX

ASSIGNEE INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY	TYPE CODE
Gist-Brocades, N.V.	Delft	N/A	N/A	NLX	03
Plant Genetic Systems, N.V.	Brussels	N/A	N/A	BEX	03

APPL-NO: 7/ 640476
DATE FILED: January 10, 1991

PARENT-CASE:
This application is a continuation-in-part of U.S. Ser. No. 466,670, filed 17 Jan. 1990, and of U.S. Ser. No. 398,519, filed 25 Aug. 1989, both now abandoned, and of U.S. Ser. No. 398,706, filed 25 Aug. 1989, now U.S. Pat. No. 5,290,690.

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	APPL-DATE
EP	88201539.9	July 15, 1988
EP	88402789.7	November 4, 1988
WO	PCT/EP89/00838	July 17, 1989
WO	PCT/EP89/00839	July 17, 1989

INT-CL: [5] C12N 9/00, C12N 9/92, C12N 11/02, C12N 15/61
US-CL-ISSUED: 435/100; 435/234, 435/827, 435/172.3, 536/23.2
US-CL-CURRENT: 435/100; 435/234, 435/827, 536/23.2
FIELD-OF-SEARCH: 435/234, 435/827, 435/172.3, 435/69.1, 435/100, 536/4.1, 536/125, 536/23.1, 536/23.2

REF-CITED:

U.S. PATENT DOCUMENTS

Search Selected Search ALL

	PAT-NO	ISSUE-DATE	PATENTEE-NAME	US-CL
☐	4567142	January 1986	Lloyd	435/94

FOREIGN PATENT DOCUMENTS

FOREIGN-PAT-NO	PUBN-DATE	COUNTRY
0275202	July 1988	EPX

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ART-UNIT: 184
PRIMARY-EXAMINER: Wax; Robert A.
ASSISTANT-EXAMINER: Jacobson; Dian C.
ATTY-AGENT-FIRM: Morrison & Foerster

ABSTRACT:

New mutant glucose isomerases are provided exhibiting improved properties under application conditions. These glucose isomerases are obtained by expression of a gene encoding said enzyme, having an amino acid sequence which differs at least in one amino acid from the wildtype glucose isomerase. Preferred mutant enzymes are those derived from Actinoplanes missouriensis glucose isomerase.

8 Claims, 38 Drawing figures

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L3: Entry 2 of 3 File: USPT Dec 27, 1994

US-PAT-NO: 5376536
DOCUMENT-IDENTIFIER: US 5376536 A

TITLE: Glucose isomerase enzymes and their use

DATE-ISSUED: December 27, 1994

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Quax; Wilhemus J.	VB Voorschoten	N/A	N/A	NLX
Luiten; Rudolf G. M.	KR Leiden	N/A	N/A	NLX
Schuurhuizen; Paul W.	NT Delft	N/A	N/A	NLX
Mrabet; Nadir	Hoeilaart	N/A	N/A	BEX

US-CL-CURRENT: 435/100; 435/234, 435/827, 536/23.2

CLAIMS:

We claim:
1. A modified glucose isomerase comprising a multimeric structure, each monomeric unit of said structure having an amino acid sequence which differs from a corresponding wild-type glucose isomerase obtained from Actinoplanes missouriensis by replacement of at least one amino acid by a different amino acid, wherein said replacement is selected from the group consisting of
replacing Lys 253 by Arg 253;
replacing Gly 70 by Ser 70;
replacing Ala 73 by Ser 73;
replacing Gly 74 by Thr 74;
replacing Lys 309 by Arg 309;
replacing Lys 319 by Arg 319; and
replacing Lys 323 by Arg 323;
said replacement not altering the glucose isomerase activity, and said modified glucose isomerase exhibiting enhanced interaction resulting in enhanced resistance of said modified glucose isomerase toward covalent binding of substrate molecules and thermostability under standard application conditions as compared to said corresponding wild-type enzyme.
2. The modified glucose isomerase of claim 1 wherein Lys253 is replaced by Arg253.

3. The modified glucose isomerase of claim 1 wherein Gly70 is replaced by Ser70; Ala73 is replaced by Ser73; and Gly74 is replaced by Thr74.
4. The modified glucose isomerase of claim 1 wherein Lys309 is replaced by Arg309; or Lys319 is replaced by Arg319; or Lys323 is replaced by Arg323.
5. The modified glucose isomerase of claim 1 wherein Lys309 is replaced by Arg309; and Lys319 is replaced by Arg319; and Lys323 is replaced by Arg323.
6. The modified glucose isomerase of claim 1 in immobilized form.
7. A method to produce fructose syrup, which method comprises contacting a preparation containing glucose with the glucose isomerase of claim 1 under conditions effective to convert a desired amount of glucose to fructose.
8. The modified glucose isomerase according to claim 1 wherein the multimeric structure is a tetramer.

WEST

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L3: Entry 1 of 3 File: USPT Jan 24, 1995

US-PAT-NO: 5384257
DOCUMENT-IDENTIFIER: US 5384257 A

TITLE: Glucose isomerases with an altered pH optimum

DATE-ISSUED: January 24, 1995

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Lambeir; Anne-Marie	Heverlee	N/A	N/A	BEX
Lasters; Ignace	Antwerp	N/A	N/A	BEX
Mrabet; Nadir	Hoeilaart	N/A	N/A	BEX
Quax; Wilhelmus J.	Voorschoten	N/A	N/A	NLX
Van der Laan; Jan M.	Groningen	N/A	N/A	NLX
Misset; Onno	Delft	N/A	N/A	NLX

ASSIGNEE INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY	TYPE CODE
Gist-brocades, N.V.	Delft	N/A	N/A	NLX	03
Plant Genetics System, NV	Brussels	N/A	N/A	BEX	03

DISCLAIMER DATE: 20110510

APPL-NO: 8/ 112703
DATE FILED: August 26, 1993

PARENT-CASE:
This application is a continuation, of application Ser. No. 07/637,399 filed Jan. 4, 1991.

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	APPL-DATE
EP	90200030	January 4, 1990
EP	90200037	January 4, 1990

INT-CL: [6] C12N 13/61, C12N 9/92, C12N 15/70, C12N 15/74
US-CL-ISSUED: 435/234; 435/69.1, 435/172.3, 435/252, 435/3257.33, 536/23.2, 935/10, 935/14, 935/60, 935/72, 935/75
US-CL-CURRENT: 435/234; 435/252.3, 435/252.33, 435/69.1, 536/23.2
FIELD-OF-SEARCH: 435/234, 435/69.1, 435/172.3, 435/252.3, 435/252.33, 435/320.1, 435/23.2

REF-CITED:

U.S. PATENT DOCUMENTS

Search Selected Search ALL

	PAT-NO	ISSUE-DATE	PATENTEE-NAME	US-CL
☐	4609625	September 1986	Keyes et al.	435/176
☐	4894331	June 1990	Ratzkin et al.	435/94
☐	5041378	August 1991	Drummond et al.	435/234

FOREIGN PATENT DOCUMENTS

FOREIGN-PAT-NO	PUBN-DATE	COUNTRY
90-0196	January 1990	WOX

OTHER PUBLICATIONS

Collyer, C. A., et al., Mar. 1990, Journal of Molecular Biology, 212(1): 211-235.
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Batt, C. A., et al., Jan. 1990, Proceedings of the National Academy of Sciences, U.S.A., 87: 618-622.
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Zvelebil, M. J. J. M., et al., 1988, Protein Engineering, 2(2): 127-138.
Farber, G. K., et al., 1988, Proceedings of the National Academy of Sciences, U.S.A., 85: 112-115.
Farber, G. K., et al., 1987, Protein Engineering, 1:467-469.
Henrik, K., et al., 1987, Protein Engineering, 1: 459-460.
Saari, G. C., et al., 1987, Journal of Bacteriology, 169(2): 612-618.
Sternberg, M. J. E., et al., 1987, Nature, 330: 86-88.
Wells, J. A., et al., 1987, Proceedings of the National Academy of Sciences, U.S.A., 84: 1219-1223.
Russel, A. J., et al., 1987, Journal of Molecular Biology 193: 803-813.
Thomas, P. G., et al., 1987, Nature, 318: 375-376.
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Rey et al., "Structure Analysis of the 2.8 .ANG. Model of Xylose Isomerase From Actinoplanes missouriensis", Proteins 4:165-172 (1987).
Henrick et al., "Comparison of backbone structures of glucose isomerase from Streptomyces and Arthrobacter", Protein Eng. 1:467-475 (1987).
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R. van Tilbert, Thesis entitled "Engineering Aspects of Biocatalysts in Industrial Starch Conversion Technology", Delftse Universitaire Pers (1983).

ART-UNIT: 184
PRIMARY-EXAMINER: Wax; Robert A.
ASSISTANT-EXAMINER: Moore; William W.
ATTY-AGENT-FIRM: Morrison & Foerster

ABSTRACT:

A method for selecting amino acid residues is disclosed which upon replacement will give rise to an enzyme with an altered pH optimum. The method is specific for metalloenzymes which are inactivated at low pH due to the dissociation of the metal ions. The method is based on altering the pK.sub.a of the metal coordinating ligands or altering the K.sub.ass for the metal binding. New glucose isomerases with an altered pH optimum are provided according to this method. These altered properties enable starch degradation to be performed at lower pH values.

15 Claims, 24 Drawing figures

L3: Entry 1 of 3 File: USPT Jan 24, 1995

US-PAT-NO: 5384257
DOCUMENT-IDENTIFIER: US 5384257 A

TITLE: Glucose isomerases with an altered pH optimum

DATE-ISSUED: January 24, 1995

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Lambeir; Anne-Marie	Heverlee	N/A	N/A	BEX
Lasters; Ignace	Antwerp	N/A	N/A	BEX
Mrabet; Nadir	Hoeilaart	N/A	N/A	BEX
Quax; Wilhelmus J.	Voorschoten	N/A	N/A	NLX
Van der Laan; Jan M.	Groningen	N/A	N/A	NLX
Misset; Onno	Delft	N/A	N/A	NLX

US-CL-CURRENT: 435/234; 435/252.3, 435/252.33, 435/69.1, 536/23.2

CLAIMS:

We claim:
1. A substantially pure, recombinantly produced, modified procaryotic glucose isomerase containing a modification wherein at least one and no more than four amino acid residues from the corresponding naturally occurring glucose isomerase within a sphere of 15 .ANG. of a bivalent metal cation coordination site is replaced by a more negatively charged amino acid, which modified glucose isomerase exhibits an altered pH/activity profile wherein at least the acidic part of the pH/activity profile is shifted to a higher pH value and wherein said modified glucose isomerase retains glucose isomerase activity.
2. The modified glucose isomerase of claim 1 wherein the corresponding naturally-occurring glucose isomerase is derived from a microorganism of the order Actinomycetales.
3. The modified glucose isomerase of claim 2 wherein the Actinomycetales microorganism is Actinoplanes missouriensis.
4. The modified glucose isomerase of claim 1 wherein said at least one amino acid of the corresponding naturally-occurring glucose isomerase replaced is selected from the group consisting of:
Ala5, Phe11, Leu15, Trp20, Gln21, Arg23, Ala25, Phe26, Asp28, Ala29, Gly47, Tyr49, Thr52, Phe53, His54, Asp56, Asp57, Phe61, Ile85, Met88, Phe94, Thr95, Phe104, Gln122, Thr133, Leu134, Val135, Ala143, Tyr145, Tyr158, Asn163, Ser169, Glu181, Asn185, Glu186, Gly189, Ile191, Pro194, His198, Gln204, Leu211, Phe212, Asn215, Glu217, Thr218, His220, Glu221, Gln222, Ser224, Asn225, Leu226, Phe228, Thr229, Gly231, Leu236, His238, His243, Asp245, Asn247, His250, Phe254, Asp255, Gln256, Asp275, Leu258, Val259, Phe260, His262, Leu271, Tyr285, Asp286, His290, Asp292, Tyr293, Lys294, Thr298, Glu299, Trp305, Ala310, Met314, Val380, and Asn383, said positions referring to the wildtype glucose isomerase of Actinopolanes missouriensis,
and wherein said amino acid replaced is an amino acid of the wildtype glucose isomerase of Actinoplanes missouriensis or is an amino acid at a corresponding position in a homologous glucose isomerase.
5. The modified glucose isomerase of claim 4 wherein said replaced amino acid is selected from the group consisting of:
Ala5, Leu15, Gln21, Arg23, Ala25, Phe26, Asp28, Ala29, Gly47, Tyr49, Thr52, Asp56, Phe61, Ile85, Met88, Thr95, Phe104, Gln122, Thr133, Leu134, Ala143, Tyr145, Tyr158, Asn163, Ser169, Asn185, Gly189, Ile191, Pro194, His198, Gln204, Leu211,

Phe212, Thr218, Gln222, Ser224, Asn225, Leu226, Phe228, Thr229, Gly231, Leu236, His238, His243, His250, Phe254, Gln256, Asp257, Leu258, Val259, His262, Leu271, Tyr285, Asp286, His290, Tyr293, Lys294, Thr298, Glu299, Trp305, Ala310, Met314, Val380, and Asn383,
said positions referring to the wildtype glucose isomerase of Actinopolanes missouriensis,
and wherein said amino acid replaced is an amino acid of the wildtype glucose isomerase of Actinoplanes missouriensis or is an amino acid at a corresponding position in a homologous glucose isomerase.

6. The modified glucose isomerase of claim 5 wherein said replaced amino acid residue is selected from the group consisting of:
Ala25, Arg23, Gly47, Tyr49, Thr52, Phe61, Ile85, Thr95, Gln122, Thr133, Tyr145, Tyr158, Gly189, Ile191, Gln204, Thr218, Gln222, Leu226, Thr229, Gly231, Leu236, Gln256, Leu258, Try293, Lys294 and Val380,
said positions referring to the wildtype glucose isomerase of Actinopolanes missouriensis,
and wherein said amino acid replaced is an amino acid of the wildtype glucose isomerase of Actinoplanes missouriensis or is an amino acid at a corresponding position in a homologous glucose isomerase.

7. The modified glucose isomerase of claim 6 wherein said amino acid replacement is selected from the group consisting of:
R23Q, H54N, T95D, H290N, and K294Q
said positions referring to the wild-type glucose isomerase of Actinoplanes missouriensis, or
wherein said amino acid replaced is an amino acid at a corresponding position in a homologous glucose isomerase.

8. The modified glucose isomerase of claim 7 wherein said amino acid replacement is R23Q.

9. The modified glucose isomerase of claim 7 wherein said amino acid replacement is H54N.

10. The modified glucose isomerase of claim 7 wherein said amino acid replacement is T95D.

11. The modified glucose isomerase of claim 7 wherein said amino acid replacement is H290N.

12. The modified glucose isomerase of claim 7 wherein said amino acid replacement is K294Q.

13. A method to produce a modified glucose isomerase of claim 1, which process comprises:
mutating a DNA sequence encoding a wildtype glucose isomerase at selected nucleotide positions;
cloning the mutated sequence into an expression vector in such a manner that the DNA sequence can be expressed;
transforming a host organism or cell with the vector;
culturing the host organism or cell; and
isolating the modified glucose isomerase from the culture.

14. A process for producing the modified glucose isomerase of claim 1 which comprises culturing a host organism transformed with a mutated glucose isomerase DNA cloned into an expression vector in such a manner that the DNA sequence can be expressed under conditions which favor expression and
isolating the modified glucose isomerase from the culture.

15. A method for altering the pH specificity of a procaryotic glucose isomerase wherein at least the acidic part of the pH/activity profile is shifted to a higher pH value and wherein said modified glucose isomerase retains glucose isomerase activity a more negatively charged amino acid
which method comprises substituting for at least one and no more than four amino acid residues from the corresponding naturally occurring glucose isomerase within a sphere of 15 .ANG. of a bivalent metal cation coordination site.